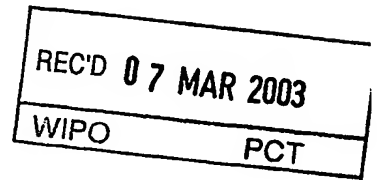




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Patentanmeldung Nr. Patent application No. Demande de brevet n°

02250289.2

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**Blatt 2 der Bescheinigung  
Sheet 2 of the certificate  
Page 2 de l'attestation**

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## POLYMER CONJUGATES

The present invention relates to conjugates of biologically active compounds, preferably therapeutically active compounds, with polymeric moieties having low polydispersity, as well as controlled polymerisation processes for producing the conjugates.

The field of polymer therapeutics involves provision of conjugates of therapeutically active compounds and polymers of various types. Selection of the class of polymer, and its properties, allows control of delivery of the therapeutic to a target within the body of the patient to be treated. Targeting may be active, by providing the conjugate with a specific label, capable of binding to complementary molecules in the target region, or passive, by controlling the molecular weight of the polymer. Conjugation of a therapeutically active compound to a polymer may control the solubility of the therapeutically active compound, its stability in the circulation, its rate of removal by the kidneys and/or liver, as well as its immunogenicity.

One example of a polymer therapeutic is the compound PK-1, a conjugate of doxorubicin with hydroxypropylmethacrylamide copolymer. The drug is conjugated to side chains on the polymer by an oligopeptidyl linker.

In WO-A-01/18080, controlled radical polymerisation processes are used to form the polymers from monomers having reactive groups, which may, after polymerisation, be used to conjugate to therapeutic ligands. The low polydispersity polymers formed by the controlled radical polymerisation processes are said to have particular advantages for therapeutic use.

Non-polymeric groups may also be conjugated to biologically active molecules, for instance therapeutically active molecules, to increase their water solubility, as well as control their stability in serum/circulation, especially for controlling drug delivery for orally administered compounds. In WO-A-9843676, US-A-5888990 and US-A-6127349, a variety of therapeutically active compounds, including compounds which are water-insoluble, are conjugated to zwitterionic groups, to control their solubility or stability in serum. The zwitterionic groups may be phosphocholine groups. The synthetic method for conjugating the phosphocholine groups generally

involves reaction of an alcohol with 2-chloro-2-oxo-1,3,2-dioxaphospholane, followed by a ring opening amination with trimethylamine. Other reactions involve conjugation of an alcohol having a PC group, with the carboxylic group of a therapeutically active using a carbodiimide to form an ester conjugate.

5 In our earlier publication WO-A-9315775, ethylenically unsaturated monomers including zwitterionic groups, for instance 2-methacryloyloxy-2'-ethyltrimethylammoniumphosphate inner salt (MPC), are polymerised by a graft polymerisation process onto carbohydrate substrates. The substrate

10 may be soluble, for instance a water soluble cellulose derivative. Polymerisation involves generation of a free radical at a hydroxyl group on the substrate by contact with cerium IV ions. The zwitterionic treated compounds were then used at surfaces of membranes to improve haemocompatibility.

15 In our earlier application, not published at the priority date hereof, PCT/GB01/04432, atom transfer radical polymerisation is carried out using monomers including zwitterionic monomers, for instance MPC. The product polymers may be used as matrices for drug delivery, for instance block copolymers may form micelles, useful as drug delivery vehicles. There is no  
20 suggestion that covalent conjugation of the drug molecule to the polymer should be considered.

Haddleton, in a paper presented at a meeting of the Society Chemical Industry London 1999, and in slides reproduced on [www.warwick.ac.uk/fac/sci/chemistry/polymers/presentations](http://www.warwick.ac.uk/fac/sci/chemistry/polymers/presentations), e.g.

25 "Functional polymers in everyday life" describes atom transfer radical polymerisation using initiators comprising biologically active molecules including carbohydrates, especially ribose moieties of nucleosides or steroids. The monomers are ethylenically unsaturated monomers which may form water soluble products. The monomers may include polyethylene glycol moieties (polyethyleneglycol methacrylate), as well as cationic groups,  
30 such as acrylic ester compounds having amine substituents in the alkyl groups. Suitable initiators are formed by acylating an alcohol derivative of

the biologically active molecule with a reactive acid compound having an  $\alpha$  halogen substituent, preferably at a tertiary carbon atom.

The present invention provides polymerisation process in which ethylenically unsaturated monomers including a zwitterionic monomer of the

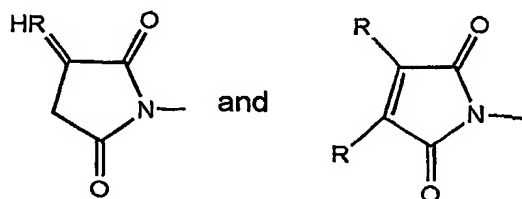
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general formula I



in which Y is an ethylenically unsaturated group selected from  $\text{H}_2\text{C}=\text{CR}-\text{CO}-\text{A}-$ ,  $\text{H}_2\text{C}=\text{CR}-\text{C}_6\text{H}_4-\text{A}^1-$ ,  $\text{H}_2\text{C}=\text{CR}-\text{CH}_2\text{A}^2$ ,  $\text{R}^2\text{O}-\text{CO}-\text{CR}=\text{CR}-\text{CO}-\text{O}$ ,  $\text{RCH}=\text{CH}-\text{CO}-\text{O}-$ ,  $\text{RCH}=\text{C}(\text{COOR}^2)\text{CH}_2-\text{CO}-\text{O}$ ,

10



15

A is  $-\text{O}-$  or  $\text{NR}^1$ ;

$\text{A}^1$  is selected from a bond,  $(\text{CH}_2)_n\text{A}^2$  and  $(\text{CH}_2)_n\text{SO}_3^-$  in which n is 1 to 12;

$\text{A}^2$  is selected from a bond,  $-\text{O}-$ ,  $\text{O}-\text{CO}-$ ,  $\text{CO}-\text{O}$ ,  $\text{CO}-\text{NR}^1-$ ,  $-\text{NR}^1-\text{CO}$ ,  $\text{O}-\text{CO}-\text{NR}^1-$ ,  $\text{NR}^1-\text{CO}-\text{O}-$ ;

20

R is hydrogen or  $\text{C}_{1-4}$  alkyl;

$\text{R}^1$  is hydrogen,  $\text{C}_{1-4}$  alkyl or  $\text{BX}$ ;

$\text{R}^2$  is hydrogen or  $\text{C}_{1-4}$  alkyl;

B is a bond, or a straight branched alkanediyl, alkylene oxaalkylene, or alkylene (oligooxaalkylene) group, optionally containing one or more

25

fluorine substituents;

X is a zwitterionic group

are polymerised by a living radical polymerisation process in the presence of an initiator, and a catalyst;

in which the initiator comprises a biologically active moiety and a

30

radical forming moiety.

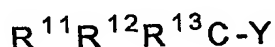
The initiator may be any compound which has a suitable radical forming moiety, which may be a moiety from which a halogen atom is

removed, or from which a radical group is removed, for instance NO-containing groups, to leave a radical at which polymerisation of the ethylenically unsaturated monomers may initiate and propagate. The radical forming moiety may comprise a halogen atom, preferably attached to a secondary or, more preferably, tertiary carbon atom, in turn joined to electron withdrawing groups such as carbonyl, sulphonyl, phosphoryl or aryl group. A halogen atom is preferably bromine but may alternatively be chlorine or iodine.

---

Preferably the radical initiator is of the general formula-V

10



V

where:

Y is selected from the group consisting of Cl, Br, I,  $OR^{10}$ ,  $SR^{14}$ ,  $SeR^{14}$ ,  $OP(=O)R^{14}$ ,  $OP(=O)(OR^{14})_2$ ,  $O-N(R^{14})_2$  and  $S-C(=S)N(R^{14})_2$ , where  $R^{10}$  is alkyl of from 1 to 20 carbon atoms in which each of the hydrogen atoms may be independently replaced by halide,  $R^{14}$  is aryl or a straight or branched  $C_1$ - $C_{20}$  alkyl group, and where an  $N(R^{14})_2$  group is present, the two  $R^{14}$  groups may be joined to form a 5- or 6-membered heterocyclic ring;

$R^{11}$  and  $R^{12}$  are each independently selected from the group consisting of H, halogen,  $C_1$ - $C_{20}$  alkyl,  $C_3$ - $C_8$  cycloalkyl,  $C(=O)R^{15}$ ,  $C(=O)NR^{16}R^{17}$ ,  $COCl$ ,  $OH$ ,  $CN$ ,  $C_2$ - $C_{20}$  alkenyl, oxiranyl, glycidyl, aryl, heterocyclyl, aralkyl and aralkenyl, in any of which the alkyl, alkenyl or aryl, heterocyclyl or cycloalkyl groups there may be from 1 to 3 substituents selected from the group consisting of hydrogen, hydroxy  $C_1$ - $C_4$  alkoxy, acyloxy, aryl, heterocyclyl,  $C(=O)R^{15}$ ,  $C(=O)NR^{16}R^{17}$ ,  $-CR^{12}R^{13}Y$ ,  $CR^{11}R^{12}Y$ , oxiranyl and glycidyl;

where  $R^{15}$  is alkyl of from 1 to 20 carbon atoms, alkoxy of from 1 to 20 carbon atoms, oligo(alkoxy) in which each alkoxy group has 1 to 3 carbon atoms, aryloxy or heterocyclyloxy any of which groups may have substituents selected from optionally substituted alkoxy, oligoalkoxy, amino (including mono-- and di-alkyl amino and trialkyl ammonium, which alkyl

groups, in turn may have substituents selected from acyl, acyloxy, alkoxy, alkoxycarbonyl, alkenoxycarbonyl, aryl and hydroxy), and hydroxyl groups;

$R^{16}$  and  $R^{17}$  are independently H or alkyl of from 1 to 20 carbon atoms which alkyl groups, in turn may have substituents selected from alkoxy, acyl, acyloxy, alkoxycarbonyl, alkenoxycarbonyl, aryl and hydroxy, or  $R^{16}$  and  $R^{17}$  may be joined together to form an alkanediyl group of from 2 to 5 carbon atoms, thus forming a 3- to 6-membered ring; and

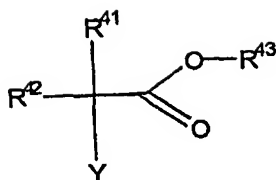
$R^{13}$  is selected from the group consisting of biologically active group-substituted alkyl, cycloalkyl,  $-\text{COR}^{15}$ ,  $-\text{CONR}^{16}\text{R}^{17}$ , alkenyl, aryl, heterocyclyl, aralkyl and aralkenyl groups, in any of which the alkyl, alkenyl or aryl, heterocyclyl or cycloalkyl groups there may be from 1 to 3 substituents selected from the group consisting of hydrogen, hydroxy  $\text{C}_1\text{-C}_4$  alkoxy, acyloxy, aryl, heterocyclyl,  $\text{C}(=\text{O})\text{R}^{15}$ ,  $\text{C}(=\text{O})\text{NR}^{16}\text{R}^{17}$ ,  $-\text{CR}^{12}\text{R}^{13}\text{Y}$ ,  $\text{CR}^{11}\text{R}^{12}\text{Y}$ , oxiranyl and glycidyl where  $R^{15}$ ,  $R^{16}$  and  $R^{17}$  are groups as defined above for  $R^{11}$  and  $R^{12}$  with the biologically active group substituted on an alkyl cycloalkyl, alkenyl aryl or heterocyclyl group.

At least one of the groups  $R^{11}$ ,  $R^{12}$  and  $R^{13}$  should be electron withdrawing, to stabilise the radical formed upon removal of Y. Suitable electron-withdrawing groups are mentioned above. Preferably one of  $R^{11}$ ,  $R^{12}$  and  $R^{13}$ , usually  $R^{13}$ , comprises the electron-withdrawing group. More preferably  $R^{13}$  is an aryl group or is a group  $\text{COR}^{15}$  in which  $R^{15}$  is alkoxy. Preferably one of  $R^{11}$  and  $R^{12}$  is H and the other is methyl.

Since any of  $R^{11}$ ,  $R^{12}$  and  $R^{13}$  may comprise a substituent  $\text{CR}^{12}\text{R}^{13}$  or  $\text{CR}^{11}\text{R}^{12}\text{Y}$ , the initiator may be di-, oligo- or poly- functional. Preferably it is a mono-functional initiator.

The biologically active moiety is preferably a pharmacologically or diagnostically active moiety, i.e. the pharmacological or diagnostic activity may be evident in the polymerised product or after cleavage of the or part of the polymeric component of the product *in vivo*, after administration. It may be derived from an alcohol, amine, carboxylic acid or other functional (derivatisable) compound which is biologically active (the base compound).

Most preferably the initiator is a compound of general formula VI



VI

where  $R^{41}$  and  $R^{42}$  are independently selected from hydrogen, straight, branched and cyclic alkyl, aryl, aralkyl, hydroxy-alkyl and acyloxyalkyl.

$R^{43}$  is a biologically active moiety;

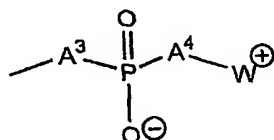
$Y$  is a radical forming group or a halogen atom.

Preferably  $R^{41}$  and  $R^{42}$  are not both hydrogen, more preferably each is methyl, or one is hydrogen and one is methyl.

$R^{43}$  is preferably derived from a pharmaceutically or diagnostically active alcohol  $R^{43}OH$ , for instance in which  $R^{43}OH$  is a carbohydrate such as a sugar.

Another example of a group  $R^{43}$  is a group  $R^{44}OL$  derived from  $R^{44}OLOH$  in which an alcohol  $R^{44}$  is derived from a pharmaceutically or diagnostically active compound  $R^{44}AH$  where  $A$  is a divalent moiety selected from the group consisting of  $O$ ,  $NR^{45}$  ( $R^{45}$  is  $H$  or lower alkyl),  $COO$  and  $CONR^{45}$ , and  $L$  is a divalent linker.  $L$  may be an oligopeptide based linker.

Preferably  $X$  is an ammonium, phosphonium, or sulphonium phosphate or phosphonate ester zwitterionic group, more preferably a group of the general formula II



II

in which the moieties  $A^3$  and  $A^4$ , which are the same or different, are  $-O-$ ,  $-S-$ ,  $-NH-$  or a valence bond, preferably  $-O-$ , and  $W^+$  is a group comprising an ammonium, phosphonium or sulphonium cationic group and a group linking the anionic and cationic moieties which is preferably a  $C_{1-12}$ -alkanediyl group,



preferably in which  $W^+$  is a group of formula  
 $-W^1-N^+R^3_3$ ,  $-W^1-P^+R^4_3$ ,  $-W^1-S^+R^4_2$  or  $-W^1-Het^+$  in which:

$W^1$  is alkanediyl of 1 or more, preferably 2-6 carbon atoms optionally  
 containing one or more ethylenically unsaturated double or triple bonds,  
 5 disubstituted-aryl (arylene), alkylene arylene, arylene alkylene, or alkylene  
 aryl alkylene, cycloalkanediyl, alkylene cycloalkyl, cycloalkyl alkylene or  
 alkylene cycloalkyl alkylene, which group  $W^1$  optionally contains one or more  
 fluorine substituents and/or one or more functional groups; and

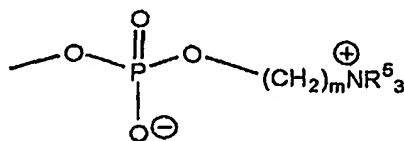
either the groups  $R^3$  are the same or different and each is hydrogen  
 10 or alkyl of 1 to 4 carbon atoms, preferably methyl, or aryl, such as phenyl, or  
 two of the groups  $R^3$  together with the nitrogen atom to which they are  
 attached form an aliphatic heterocyclic ring containing from 5 to 7 atoms, or  
 the three groups  $R^3$  together with the nitrogen atom to which they are  
 attached form a fused ring structure containing from 5 to 7 atoms in each  
 15 ring, and optionally one or more of the groups  $R^3$  is substituted by a  
 hydrophilic functional group, and

the groups  $R^4$  are the same or different and each is  $R^3$  or a group  
 $OR^3$ , where  $R^3$  is as defined above; or

Het is an aromatic nitrogen-, phosphorus- or sulphur-, preferably  
 20 nitrogen-, containing ring, for example pyridine.

Monomers in which X is of the general formula in which  $W^+$  is  
 $W^1N^+R^3_3$  may be made as described in our earlier specification WO-A-  
 9301221. Phosphonium and sulphonium analogues are described in WO-A-  
 9520407 and WO-A-9416749.

25 Generally a group of the formula II has the preferred general formula  
 III

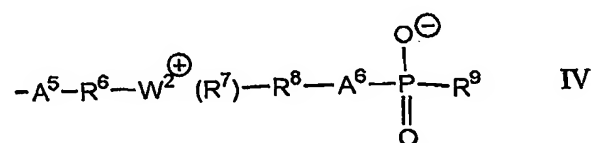


III

where the groups  $R^5$  are the same or different and each is hydrogen or  $C_{1-4}$  alkyl, and  $m$  is from 1 to 4, in which preferably the groups  $R^5$  are the same preferably methyl.

In phosphobetaine based groups, X may have the general formula IV

5




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in which  $A^5$  is a valence bond,  $-O-$ ,  $-S-$  or  $-NH-$ , preferably  $-O-$ ;

10

$R^6$  is a valence bond (together with  $A^5$ ) or alkanediyl,  $-C(O)$ alkylene- or  $-C(O)NH$  alkylene preferably alkanediyl, and preferably containing from 1 to 6 carbon atoms in the alkanediyl chain;

$W^2$  is S,  $PR^7$  or  $NR^7$ ;

15

the or each group  $R^7$  is hydrogen or alkyl of 1 to 4 carbon atoms or the two groups  $R^7$  together with the heteroatom to which they are attached form a heterocyclic ring of 5 to 7 atoms;

$R^8$  is alkanediyl of 1 to 20, preferably 1 to 10, more preferably 1 to 6 carbon atoms;

$A^6$  is a bond,  $NH$ , S or O, preferably O; and

20

$R^9$  is a hydroxyl,  $C_{1-12}$  alkyl,  $C_{1-12}$  alkoxy,  $C_{7-18}$  aralkyl,  $C_{7-18}$  -aralkoxy,  $C_{6-18}$  aryl or  $C_{6-18}$  aryloxy group.

Monomers comprising a group of the general formula IV may be made by methods as described in JP-B-03-031718, in which an amino substituted monomer is reacted with a phospholane.

25

In compounds comprising a group of the general formula IV, it is preferred that

$A^5$  is a bond;

$R^6$  is a  $C_{2-6}$  alkanediyl;

$W^2$  is  $NR^7$ ;

30

each  $R^7$  is  $C_{1-4}$  alkyl;

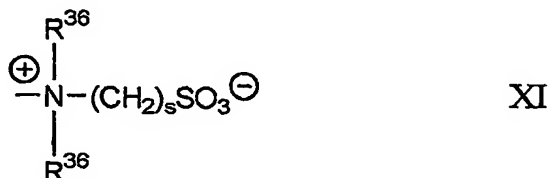
$R^8$  is  $C_{2-6}$  alkanediyl;

$A^6$  is O; and

R<sup>9</sup> is C<sub>1-4</sub> alkoxy.

Alternatively X may be a zwitterion in which the anion comprises a sulphate, sulphonate or carboxylate group.

One example of such a group is a sulphobetaine group, of the general  
5 formula XI

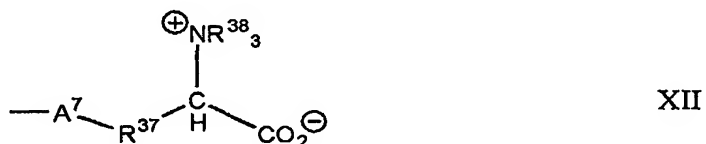


10 where the groups R<sup>36</sup> are the same or different and each is hydrogen or C<sub>1-4</sub> alkyl and s is from 2 to 4.

Preferably the groups R<sup>36</sup> are the same. It is also preferable that at least one of the groups R<sup>36</sup> is methyl, and more preferable that the groups R<sup>36</sup> are both methyl.

15 Preferably s is 2 or 3, more preferably 3.

Another example of a zwitterionic group having a carboxylate group is an amino acid moiety in which the alpha carbon atom (to which an amine group and the carboxylic acid group are attached) is joined through a linker group to the backbone of the biocompatible polymer. Such groups may be  
20 represented by the general formula XII



25 in which A<sup>7</sup> is a valence bond, -O-, -S- or -NH-, preferably -O-,

R<sup>37</sup> is a valence bond (optionally together with A<sup>7</sup>) or alkanediyl, -C(O)alkylene- or -C(O)NHalkylene, preferably alkanediyl and preferably containing from 1 to 6 carbon atoms; and

the groups R<sup>38</sup> are the same or different and each is hydrogen or alkyl  
30 of 1 to 4 carbon atoms, preferably methyl, or two or three of the groups R<sup>38</sup>, together with the nitrogen to which they are attached, form a heterocyclic ring of from 5 to 7 atoms, or the three group R<sup>38</sup> together with the nitrogen

atom to which they are attached form a fused ring heterocyclic structure containing from 5 to 7 atoms in each ring.

Another example of a zwitterion having a carboxylate group is a carboxy betaine  $-N^+(R^{39})_2(CH_2)_rCOO^-$  in which the  $R^{39}$  groups are the same or different and each is hydrogen or  $R_{1-4}$  alkyl and  $r$  is 2 to 6, preferably 2 or 3.

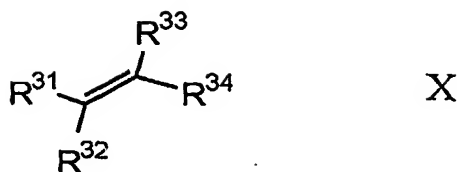
In the zwitterionic monomer of the general formula I it is preferred that the ethylenic unsaturated group  $Y$  is  $H_2C=CR-CO-A-$ . Such acrylic moieties are preferably methacrylic, that is in which  $R$  is methyl, or acrylic, in which  $R$  is hydrogen. Whilst the compounds may be (meth)acrylamido compounds (in which  $A$  is  $NR^1$ ), in which case  $R^1$  is preferably hydrogen, or less preferably, methyl, most preferably the compounds are esters, that is in which  $A$  is  $O$ .

In monomers of the general formula I, especially where  $Y$  is the preferred (alk)acrylic group,  $B$  is most preferably an alkanediyl group. Whilst some of the hydrogen atoms of such group may be substituted by fluorine atoms, preferably  $B$  is an unsubstituted alkanediyl group, most preferably a straight chain group having 2 to 6 carbon atoms.

A particularly preferred zwitterionic monomer is 2-methacryloyloxyethyl-2'-trimethylammonium ethyl phosphate inner salt.

In the polymerisation process, the ethylenically unsaturated monomers may further include a comonomer. Comonomers are copolymerisable with the zwitterionic monomer and are preferably selected from anionic, cationic and nonionic monomers. It is generally preferred that the monomer mixture include at least one nonionic monomer. Another class of comonomer is a cross-linking monomer having a functional group which may be cured after polymerisation to cross-link the polymer.

Examples of suitable comonomers are compounds of the general formula X



5 in which  $\text{R}^{31}$  is selected from hydrogen, halogen,  $\text{C}_{1-4}$  alkyl and groups  $\text{COOR}^2$  in which  $\text{R}^2$  is hydrogen and  $\text{C}_{1-4}$  alkyl;

$\text{R}^{32}$  is selected from hydrogen, halogen and  $\text{C}_{1-4}$  alkyl;

$\text{R}^{33}$  is selected from hydrogen, halogen,  $\text{C}_{1-4}$  alkyl and groups  $\text{COOR}^2$  provided that  $\text{R}^{31}$  and  $\text{R}^{33}$  are not both  $\text{COOR}^2$ ; and

10  $\text{R}^{34}$  is a  $\text{C}_{1-10}$  alkyl, a  $\text{C}_{1-20}$  alkoxy carbonyl, a mono-or di- ( $\text{C}_{1-20}$  alkyl) amino carbonyl, a  $\text{C}_{6-20}$  aryl (including alkaryl) a  $\text{C}_{7-20}$  aralkyl, a  $\text{C}_{6-20}$  aryloxy carbonyl, a  $\text{C}_{1-20}$  -aralkyloxy carbonyl, a  $\text{C}_{6-20}$  arylamino carbonyl, a  $\text{C}_{7-20}$  aralkyl-amino, a hydroxyl or a  $\text{C}_{2-10}$  acyloxy group, any of which may have one or more substituents selected from halogen atoms, alkoxy, oligo-alkoxy, 15 aryloxy, acyloxy, acylamino, amine (including mono and di-alkyl amino and trialkylammonium in which the alkyl groups may be substituted), carboxyl, sulphonyl, phosphoryl, phosphino, (including mono- and di- alkyl phosphine and tri-alkylphosphonium), zwitterionic, hydroxyl groups, vinyloxy carbonyl and other vinylic or allylic substituents, and reactive silyl or silyloxy groups, 20 such as trialkoxysilyl groups;

or  $\text{R}^{34}$  and  $\text{R}^{33}$  or  $\text{R}^{34}$  and  $\text{R}^{32}$  may together form  $-\text{CONR}^{35}\text{CO}$  in which  $\text{R}^{35}$  is a  $\text{C}_{1-20}$  alkyl group.

It is preferred for at least two of the groups  $\text{R}^{31}$ ,  $\text{R}^{32}$ ,  $\text{R}^{33}$  and  $\text{R}^{34}$  to be halogen or, more preferably, hydrogen atoms. Preferably  $\text{R}^{31}$  and  $\text{R}^{32}$  are 25 both hydrogen atoms. It is particularly preferred that compound of general formula X be a styrene-based or acrylic based compound. In styrene based compounds  $\text{R}^{34}$  represents an aryl group, especially a substituted aryl group in which the substituent is an amino alkyl group, a carboxylate or a sulphonate group. Where the comonomer is an acrylic type compound,  $\text{R}^{34}$  30 is an alkoxy carbonyl, an alkyl amino carbonyl, or an aryloxy carbonyl group. Most preferably in such compounds  $\text{R}^{34}$  is a  $\text{C}_{1-20}$  -alkoxy carbonyl group,

optionally having a hydroxy substituent. Acrylic compounds are generally methacrylic in which case  $R^{33}$  is methyl.

Where a comonomer is included in the polymerisation process of the invention, the molar ratio of zwitterionic monomer to comonomer is preferably in the range 1:50 to 50:1, more preferably in the range 1:10 to 10:1, more preferably in the range 1:5 to 1:1.

In the atom or group radical transfer polymerisation process the transition metal compound which comprises a component of the catalyst is

$M_t^{n+}X'_n$  where.

$M_t^{n+}$  may be selected from the group consisting of  $Cu^{1+}$ ,  $Cu^{2+}$ ,  $Fe^{2+}$ ,  $Fe^{3+}$ ,  $Ru^{2+}$ ,  $Ru^{3+}$ ,  $Cr^{2+}$ ,  $Cr^{3+}$ ,  $Mo^{2+}$ ,  $Mo^{3+}$ ,  $W^{2+}$ ,  $W^{3+}$ ,  $Mn^{2+}$ ,  $Mn^{3+}$ ,  $Mn^{4+}$ ,  $Rh^{3+}$ ,  $Rh^{4+}$ ,  $Re^{2+}$ ,  $Re^{3+}$ ,  $Co^{+}$ ,  $Co^{2+}$ ,  $Co^{3+}$ ,  $V^{2+}$ ,  $V^{3+}$ ,  $Zn^{+}$ ,  $Zn^{2+}$ ,  $Ni^{2+}$ ,  $Ni^{3+}$ ,  $Au^{+}$ ,  $Au^{2+}$ ,  $Ag^{+}$  and  $Ag^{2+}$ ;

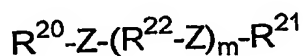
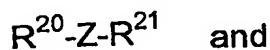
$X'$  is selected from the group consisting of halogen,  $C_1$ - $C_6$ -alkoxy,  $(SO_4)_{1/2}$ ,  $(PO_4)_{1/3}$ ,  $(R^{18}PO_4)_{1/2}$ ,  $(R^{18}_2PO_4)$ , triflate, hexafluorophosphate, methanesulphonate, arylsulphonate, CN and  $R^{19}CO_2$ , where  $R^{18}$  is aryl or a straight or branched  $C_{1-20}$  alkyl and  $R^{19}$  is H or a straight or branched  $C_1$ - $C_6$  alkyl group which may be substituted from 1 to 5 times with a halogen; and

$n$  is the formal charge on the metal ( $0 \leq n \leq 7$ ).

Preferably  $X'$  is halide, most preferably chloride or bromide. Particularly suitable transition metal compounds are based on copper or ruthenium, for instance  $CuCl$  or  $RuCl_2$ .

In the catalyst, the ligand is preferably selected from the group consisting of:

a) compounds of the formulas:



where:

$R^{20}$  and  $R^{21}$  are independently selected from the group consisting of H,  $C_1$ - $C_{20}$  alkyl, aryl, heterocyclyl and  $C_1$ - $C_6$  alkoxy,  $C_1$ - $C_4$  dialkylamino,

$C(=O)R^{22}$ ,  $C(=O)R^{23}R^{24}$  and  $A^7C(=O)R^{25}$ , where  $A^7$  may be  $NR^{26}$  or  $O$ ;  $R^{22}$  is alkyl of from 1 to 20 carbon atoms, aryloxy or heterocyclyloxy;  $R^{23}$  and  $R^{24}$  are independently  $H$  or alkyl of from 1 to 20 carbon atoms or  $R^{23}$  and  $R^{24}$  may be joined together to form an alkanediyl group of from 2 to 5 carbon atoms, thus forming a 3- to 6-membered ring;  $R^{25}$  is  $H$ , straight or branched  $C_1$ - $C_{20}$  alkyl or aryl and  $R^{26}$  is hydrogen, straight or branched;  $C_{1-20}$ -alkyl or aryl; or  $R^{20}$  and  $R^{21}$  may be joined to form, together with  $Z$ , a saturated or unsaturated ring;

$Z$  is  $O$ ,  $S$ ,  $NR^{27}$  or  $PR^{27}$ , where  $R^{27}$  is selected from the same group as  $R^{20}$  and  $R^{21}$ , and where  $Z$  is  $PR^{27}$ ,  $R^{27}$  can also  $C_1$ - $C_{20}$  alkoxy or  $Z$  may be a bond,  $CH_2$  or a fused ring, where one or both of  $R^{20}$  and  $R^{21}$  is heterocyclyl, each  $R^{22}$  is independently a divalent group selected from the group consisting of  $C_1$ - $C_8$  cycloalkanediyl,  $C_1$ - $C_8$  cycloalkanediyl, arenediyl and heterocyclylene where the covalent bonds to each  $Z$  are at vicinal positions or  $R^{22}$  may be joined to one or both of  $R^{20}$  and  $R^{21}$  to formulate a heterocyclic ring system; and

$m$  is from 1 to 6;

b)  $CO$ ;

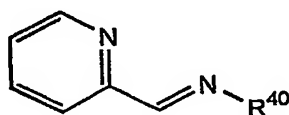
c) porphyrins and porphycenes, which may be substituted with from 1 to 6 halogen atoms,  $C_{1-6}$  alkyl groups,  $C_{1-6}$ -alkoxy groups,  $C_{1-6}$  alkoxycarbonyl, aryl groups, heterocyclyl groups, and  $C_{1-6}$  alkyl groups further substituted with from 1 to 3 halogens;

d) compounds of the formula  $R^{23}R^{24}C(C(=O)R^{25})_2$ , where  $R^{25}$  is  $C_{1-20}$  alkyl,  $C_{1-20}$  alkoxy, aryloxy or heterocyclyloxy; and each of  $R^{23}$  and  $R^{24}$  is independently selected from the group consisting of  $H$ , halogen,  $C_{1-20}$  alkyl, aryl and heterocyclyl, and  $R^{23}$  and  $R^{24}$  may be joined to form a  $C_{1-8}$  cycloalkyl ring or a hydrogenated aromatic or heterocyclic ring, of which the ring atoms may be further substituted with 1 to 5  $C_{1-6}$  alkyl groups,  $C_{1-6}$  alkoxy groups, halogen atoms, aryl groups, or combinations thereof; and

e) arenes and cyclopentadienyl ligands, where said cyclopentadienyl ligand may be substituted with from one to five methyl

groups, or may be linked through an ethylene or propylene chain to a second cyclopentadienyl ligand.

Selection of a suitable ligand is, for instance, based upon the solubility characteristics and/or the separability of the catalyst from the product polymer mixture. Generally it is catalyst to be soluble in a liquid reaction mixture, although under some circumstances it may be possible to immobilise the catalyst, for instance on a porous substrate. For the preferred process, which is carried out in the liquid phase, the ligand is soluble in a liquid phase. The ligand is generally a nitrogen-containing ligand. The preferred ligand may be a compound including a pyridyl group and an imino moiety, such as bipyridine, or



VII

where  $R^{40}$  is a suitable alkyl group, the substituent being variable and adaptable to confer desired solubility characteristics or may be triphenylphosphine or 1,1,4,7,10,10-hexamethyl-triethylene tetramine.

Such nitrogen-containing ligands are usefully used in combination with copper (I) chloride, copper (I) bromide or ruthenium chloride transition metal compounds as part of the catalyst.

The living radical polymerisation process of the invention is preferably carried out to achieve a degree of polymerisation in the range 2 to 100. Preferably the degree of polymerisation is in the range 5 to 50, more preferably in the range 10 to 25. In the preferred group or atom transfer radical polymerisation technique, the degree of polymerisation is directly related to the initial ratios of initiator to monomer. Preferably the ratio is in the range 1:(2 to 100), more preferably in the range of 1:(5 to 50), most preferably in the range 1:(10 to 25).

The ratio of metal compound and ligand in the catalyst should be approximately stoichiometric, based on the ratios of the components when the metal ion is fully complexed. The ratio should preferably be in the range



1:(0.5 to 2) more preferably in the range 1:(0.8:1.25). Preferably the range is about 1:1.

In the process, the catalyst may be used in amounts such that a molar equivalent quantity as compared to the level of initiator is present. However, since catalyst is not consumed in the reaction, it is generally not essential to include levels of catalyst as high as of initiator. The ratio of catalyst (based on transition metal compound) to initiator is preferably in the range 1:(1 to 50), more preferably in the range 1:(1 to 10).

Whilst the polymerisation reaction may be carried out in the gaseous phase, it is more preferably carried out in the liquid phase. The reaction may be heterogeneous, that is comprising a solid and a liquid phase, but is more preferably homogeneous. Preferably the polymerisation is carried out in a single liquid phase. Where the monomer is liquid, it is sometimes unnecessary to include a non-polymerisable solvent. More often, however, the polymerisation takes place in the presence of a non-polymerisable solvent. The solvent should be selected having regard to the nature of the zwitterionic monomer and any comonomer, for instance for its suitability for providing a common solution containing both monomers. The solvent may comprise a single compound or a mixture of compounds.

Where the zwitterionic monomer is MPC, may be desirable to include water in the polymerisation mixture. Preferably water should be present in an amount in the range 10 to 100% by weight based on the weight of ethylenically unsaturated monomer. Preferably the total non-polymerisable solvent comprised 1 to 500% by weight based on the weight of ethylenically unsaturated monomer. It has been found that the zwitterionic monomer and water should be in contact with each other for as short a period as possible prior to contact with the initiator and catalyst. It may be desirable therefore for all the components of the polymerisation other than the zwitterionic monomer to be premixed and for the zwitterionic monomer to be added to the premix as the last additive.

It is often desired to copolymerise MPC or other zwitterionic monomer with a comonomer and/or an initiator which is insoluble in water. In such

circumstances, a solvent or co-solvent (in conjunction with water) is included to confer solubility on both MPC and the more hydrophobic monomer.

Suitable organic solvents are ethers, esters and, most preferably, alcohols.

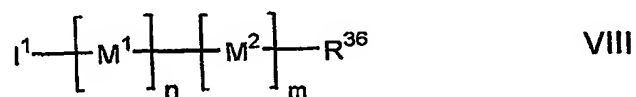
Especially where a mixture of organic solvent and water is to be used, suitable alcohols are C<sub>1-4</sub>-alkanols. Methanol is expected to be particularly suitable in the polymerisation process of the invention. Ethanol and isopropanol are likely to be useful also.

The process may be carried out at raised temperature, for instance up to 60 to 80 °C. The process may proceed sufficiently fast at ambient

temperature.

The polymerisation process of the invention should preferably be carried out so as to provide polymers of zwitterionic monomers having a polydispersity (of molecular weight) of less than 1.5, as judged by gel permeation chromatography. Polydispersities in the range 1.2 to 1.4 are preferred. Conversion rates achieved in the process should be as high as possible, for instance over 90% often over 95% or higher. It is preferred that the process be continued until a conversion level of at least 50%, or usually, at least 70% is reached.

According to a further aspect of the invention there is provided a novel compound comprising a conjugate of a biologically active moiety and a polymer group having a general formula VII:



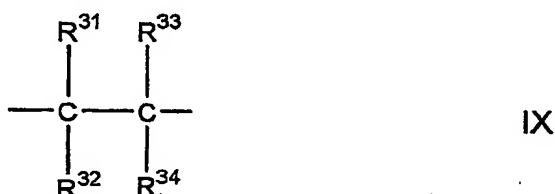
in which M<sup>1</sup> is the divalent group formed when the compound of the general formula I as defined in claim 1 is polymerised, M<sup>2</sup> is the divalent group formed when a comonomer as defined in claim 19 is polymerised, and I<sup>1</sup> is the residue of the initiator defined in claim 1 which comprises said biologically active moiety, and R<sup>36</sup> is a monofunctional group or atom which terminates the polymeric group -M<sup>1</sup><sub>n</sub>M<sup>2</sup><sub>m</sub>-, n is at least 2 and m is at least 0.

Preferably the degree of polymerisation of monomers forming residues M<sup>1</sup> is at least 5, more preferably at least 10, that is n is at least 5,

preferably at least 10. The value of  $n$  is generally no more than 100, preferably less than 50.

Groups  $M^2$  are derived from comonomer, for instance of the general formula X defined above. The groups  $M^2$  thus have the general formula IX

5



10 Where a comonomer is not included,  $n$  is 0. In the general formula defining the polymer, the groups  $M^1$  and  $M^2$  are randomly arranged. The ratio  $n:m$  represents the molar ratio of zwitterionic monomer to comonomer used in the polymerisation.

15  $R^{36}$  may include a further polymeric block, for instance, formed in a second controlled radical polymerisation step involving a further ethylenically unsaturated monomer, for instance which may have the general formula X above.

The selection of suitable monomer or monomer mixtures to form such a second block may allow an amphiphilic polymer to be formed, i.e. if the  
20 second block is hydrophobic relative to the block  $\left[ M^1 \right]_n \left[ M^2 \right]_m$ .

The present invention also covers reverse block copolymers to the type described above, that is where a first block is formed by polymerisation of monomers not including a monomer of the formula I, in the presence of the specified initiator, and a second block is formed in a second controlled  
25 radical polymerisation of monomer including zwitterionic monomer in the presence of the product of the first polymerisation step.

The degree of polymerisation and the polydispersity of the polymeric group of the conjugate of the invention produced by the polymerisation process of the first aspect of the invention, allows for control of the solubility  
30 of the biologically active compound. Where the compound is therapeutically

active, for instance, control of the nature of the polymeric group may allow for good control of delivery of the therapeutic to a target, organ, tumour or cell. The molecular weight should be high enough to minimise removal of the conjugate by the kidneys, but should be below the threshold which renders a compound hepatoselective. For instance the weight should be selected so as to take advantage of the phenomenon of enhanced permeability and retention (EPR) observed in solid tumours where the active is an anti-tumour agent for local delivery to the tumour. It may be desirable

to provide the conjugate with an active targeting moiety, for instance by

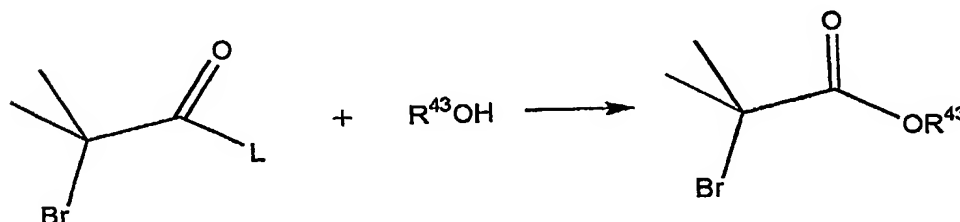
derivatising terminal groups  $R^{36}$  with a binding moiety, for instance an antigen, antibody, oligopeptide, oligonucleotide, lectin, biotin, or other member of a specific binding pair, capable of binding to another number of the binding pair localised at the target organ. The biologically active compound may be a protein or polypeptide, a carbohydrate, a nucleic acid or another drug, or may comprise a combination of such components.

The polymeric group may further provide improved stability in the circulation or in serum, or improved circulation times, as has been described for the monomeric PC-ylation in WO-A-9843676 etc. The invention is believed to be of particular utility where the compound from which the biologically active compound is derived (e.g.  $R^{44}OH$  or  $R^{43}OH$ ) has limited water solubility, for instance solubility less than in the range 0.1 to 10 mg/ml. Preferably the water solubility of the conjugate compound is at least 10 times higher, preferably at least 100 times higher than the base compound.

The invention is illustrated in the following reaction schemes.

### Reference Example 1

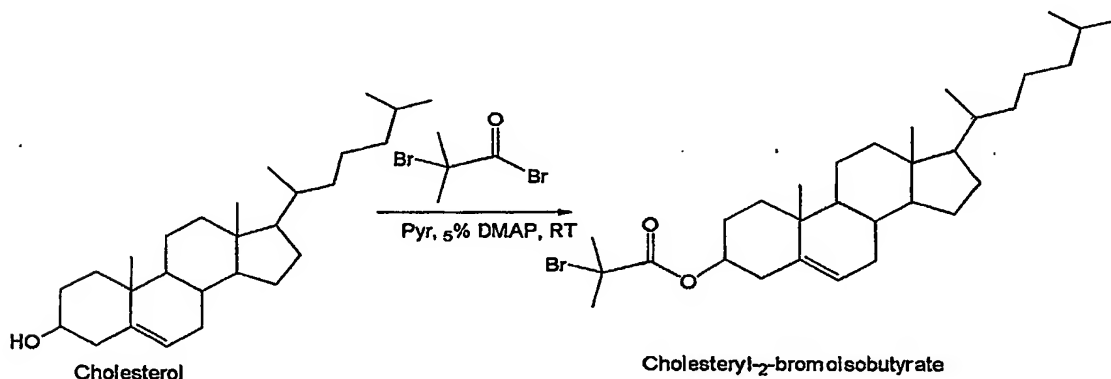
#### **General method of producing initiators**



In the above reaction, L is a leaving group. This may be for instance a halogen atom, an acyloxy group (i.e. the acylation reagent is an anhydride), or a group  $-\text{ON}=\text{C}(\text{Me})_2$ . Where the leaving group is the last mentioned compound the esterification reaction may be conducted in dioxane, at raised temperature, for instance  $60^\circ\text{C}$ .

The above general reaction is carried out with compounds in which  $\text{R}^{43}\text{OH}$  is a suitably protected carbohydrate, that is one in which all the hydroxyl groups apart from the hydroxyl group desired to be derivatised, are protected with removable groups. Conventional protecting group chemistry is appropriate. The carbohydrate may be a ribose group of a nucleoside. Alternatively  $\text{R}^{43}\text{OH}$  may be an oligosaccharide. In a carbohydrate  $\text{R}^{43}\text{OH}$  or a sugar such as glucose or galactose all other hydroxyl groups are protected during the reaction, for instance by acetylation or by protection of *gem*. diols by isopropylidene groups. Alternatively  $\text{R}^{43}\text{OH}$  may be cholesterol.

A specific initiator may be formed by the following reaction scheme:



### Example 1

Cholesteryl-2-bromoisobutyrate is used as an initiator in an atom transfer radical polymerisation process using copper (I) bromide and a 2-pyridinecarbaldehyde imine ligand, as described in US-A-6310149, using MPC monomer to form a polymer having a degree of polymerisation between 2 and 50, for instance in the range 5 to 20, using an appropriate molar ratio of initiator to monomer. The solvent is selected to provide common solubility of the initiator and the monomer, and to allow recovery of the polymer.

An alternative polymerisation uses copper (I) bromide catalyst and bi  
pyridine ligand. The solvent system in this case preferably includes water,  
or alcohol, preferably methanol, or a mixture of water and methanol.

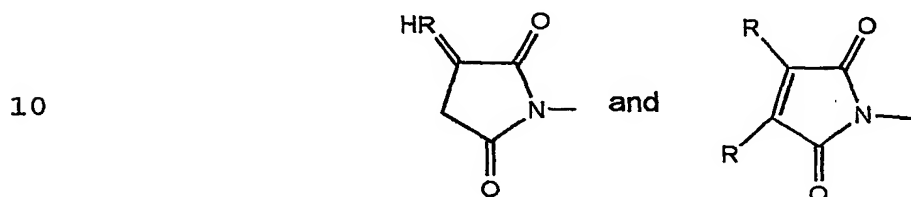
- 5 The product may be analysed by gel permeation chromatography, to  
assess the number average and weight average molecular weight and the  
polydispersity.

**CLAIMS**

1. A polymerisation process in which ethylenically unsaturated monomers including a zwitterionic monomer of the general formula I



5 in which Y is an ethylenically unsaturated group selected from  $\text{H}_2\text{C}=\text{CR}-\text{CO}-\text{A}-$ ,  $\text{H}_2\text{C}=\text{CR}-\text{C}_6\text{H}_4-\text{A}^1-$ ,  $\text{H}_2\text{C}=\text{CR}-\text{CH}_2\text{A}^2$ ,  $\text{R}^2\text{O}-\text{CO}-\text{CR}=\text{CR}-\text{CO}-\text{O}$ ,  $\text{RCH}=\text{CH}-\text{CO}-\text{O}-$ ,  $\text{RCH}=\text{C}(\text{COOR}^2)\text{CH}_2-\text{CO}-\text{O}$ ,



A is  $-\text{O}-$  or  $\text{NR}^1$ ;

15  $\text{A}^1$  is selected from a bond,  $(\text{CH}_2)_n\text{A}^2$  and  $(\text{CH}_2)_n\text{SO}_3^-$  in which n is 1 to 12;

$\text{A}^2$  is selected from a bond,  $-\text{O}-$ ,  $\text{O}-\text{CO}-$ ,  $\text{CO}-\text{O}$ ,  $\text{CO}-\text{NR}^1-$ ,  $-\text{NR}^1-\text{CO}$ ,  $\text{O}-\text{CO}-\text{NR}^1-$ ,  $\text{NR}^1-\text{CO}-\text{O}-$ ;

R is hydrogen or  $\text{C}_{1-4}$  alkyl;

$\text{R}^1$  is hydrogen,  $\text{C}_{1-4}$  alkyl or  $\text{BX}$ ;

20  $\text{R}^2$  is hydrogen or  $\text{C}_{1-4}$  alkyl;

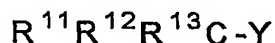
B is a bond, or a straight branched alkanediyl, alkylene oxaalkylene, or alkylene (oligooxaalkylene) group, optionally containing one or more fluorine substituents;

X is a zwitterionic group

25 are polymerised by a living radical polymerisation process in the presence of an initiator, and a catalyst;

in which the initiator comprises a biologically active moiety and a radical forming moiety.

2. A process according to claim 1 in which the initiator is a  
30 compound of general formula the radical initiator is of the general formula V



V

where:

Y is selected from the group consisting of Cl, Br, I,  $\text{OR}^{10}$ ,  $\text{SR}^{14}$ ,  $\text{SeR}^{14}$ ,  $\text{OP}(=\text{O})\text{R}^{14}$ ,  $\text{OP}(=\text{O})(\text{OR}^{14})_2$ ,  $\text{O-N}(\text{R}^{14})_2$  and  $\text{S-C}(=\text{S})\text{N}(\text{R}^{14})_2$ , where  $\text{R}^{10}$  is alkyl of from 1 to 20 carbon atoms in which each of the hydrogen atoms may  
 5 be independently replaced by halide,  $\text{R}^{14}$  is aryl or a straight or branched  $\text{C}_1$ - $\text{C}_{20}$  alkyl group, and where an  $\text{N}(\text{R}^{14})_2$  group is present, the two  $\text{R}^{14}$  groups may be joined to form a 5- or 6-membered heterocyclic ring;

$\text{R}^{11}$  and  $\text{R}^{12}$  are each independently selected from the group consisting of H, halogen,  $\text{C}_1$ - $\text{C}_{20}$  alkyl,  $\text{C}_3$ - $\text{C}_8$  cycloalkyl,  $\text{C}(=\text{O})\text{R}^{15}$ ,  
 10  $\text{C}(=\text{O})\text{NR}^{16}\text{R}^{17}$ ,  $\text{COCl}$ ,  $\text{OH}$ ,  $\text{CN}$ ,  $\text{C}_2$ - $\text{C}_{20}$  alkenyl, oxiranyl, glycidyl, aryl, heterocyclyl, aralkyl and aralkenyl, in any of which the alkyl, alkenyl or aryl, heterocyclyl or cycloalkyl groups there may be from 1 to 3 substituents selected from the group consisting of hydrogen, hydroxy  $\text{C}_1$ - $\text{C}_4$  alkoxy, acyloxy, aryl, heterocyclyl,  $\text{C}(=\text{O})\text{R}^{15}$ ,  $\text{C}(=\text{O})\text{NR}^{16}\text{R}^{17}$ ,  $-\text{CR}^{12}\text{R}^{13}\text{Y}$ ,  $\text{CR}^{11}\text{R}^{12}\text{Y}$ ,  
 15 oxiranyl and glycidyl;

where  $\text{R}^{15}$  is alkyl of from 1 to 20 carbon atoms, alkoxy of from 1 to 20 carbon atoms, oligo(alkoxy) in which each alkoxy group has 1 to 3 carbon atoms, aryloxy or heterocycloxy any of which groups may have substituents selected from optionally substituted alkoxy, oligoalkoxy, amino  
 20 (including mono-- and di-alkyl amino and trialkyl ammonium, which alkyl groups, in turn may have substituents selected from acyl, acyloxy, alkoxy, alkoxycarbonyl, alkenoxycarbonyl, aryl and hydroxy), and hydroxyl groups;

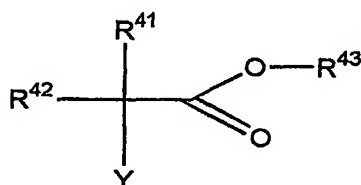
$\text{R}^{16}$  and  $\text{R}^{17}$  are independently H or alkyl of from 1 to 20 carbon atoms which alkyl groups, in turn may have substituents selected from alkoxy, acyl, acyloxy, alkoxycarbonyl, alkenoxycarbonyl, aryl and hydroxy, or  $\text{R}^{16}$  and  $\text{R}^{17}$   
 25 may be joined together to form an alkanediyl group of from 2 to 5 carbon atoms, thus forming a 3- to 6-membered ring; and

$\text{R}^{13}$  is selected from the group consisting of biologically active group-substituted alkyl, cycloalkyl,  $-\text{COR}^{15}$ ,  $-\text{CONR}^{16}\text{R}^{17}$ , alkenyl, aryl, heterocyclyl, aralkyl and aralkenyl groups, in any of which the alkyl, alkenyl or aryl,  
 30 heterocyclyl or cycloalkyl groups there may be from 1 to 3 substituents selected from the group consisting of hydrogen, hydroxy  $\text{C}_1$ - $\text{C}_4$  alkoxy,



acyloxy, aryl, heterocyclyl,  $C(=O)R^{15}$ ,  $C(=O)NR^{16}R^{17}$ ,  $-CR^{12}R^{13}Y$ ,  $CR^{11}R^{12}Y$ , oxiranyl and glycidyl where  $R^{15}$ ,  $R^{16}$  and  $R^{17}$  are groups as defined above for  $R^{11}$  and  $R^{12}$  with the biologically active group substituted on an alkyl cycloalkyl, alkenyl aryl or heterocyclyl group.

- 5            3.      A process according to claim 2 in which the initiator is a compound of general formula VI



VI

10            where  $R^{41}$  and  $R^{42}$  are independently selected from hydrogen, straight, branched and cyclic alkyl, aryl, aralkyl, hydroxy-alkyl and acyloxyalkyl.

$R^{43}$  is a biologically active moiety;

- 15            Y as defined in claim 2 and is preferably a halogen atom.

4.      A process according to claim 3 in which  $R^{41}$  and  $R^{42}$  are not both hydrogen, preferably in which either

- a)       $R^{41}$  and  $R^{42}$  are each methyl; or
- b)       $R^{41}$  is hydrogen and  $R^{42}$  is methyl.

- 20            5.      A process according to claim 1 in which the biologically active moiety is a steroid moiety, for instance a cholesterol moiety.

6.      A process according to claims 3 or claim 4 in which  $R^{43}$  is derived from a pharmaceutically or diagnostically active alcohol  $R^{43}OH$ .

- 25            7.      A process according to claim 6 in which  $R^{43}OH$  is a carbohydrate, for instance a saccharide.

8.      A process according to claim 3 or claim 4 in which  $R^{43}$  is  $R^{44}OL$ - derived from  $R^{44}OLOH$  in which  $R^{44}$  is derived from a pharmacologically or diagnostically active compound  $R^{44}AH$  where A is a divalent moiety selected from the group consisting of O,  $NR^{35}$  ( $R^{35}$  is H or lower alkyl), COO and  $CONR^{35}$ , and L is a divalent linker.
- 30

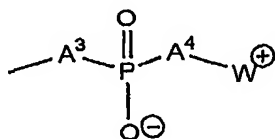
9.      A process according to claim 8 in which L is an oligo-peptide-based linker.

10. A process according to any preceding claim in which the product polymer has a molecular weight in the range 1000 to 100,000, preferably 20,000 to 50,000.

11. A process according to any preceding claim in which the product polymer has a polydispersity less than 1.5, preferably less than 1.2.

12. A process according to any preceding claim in which X is an ammonium, phosphonium, or sulphonium phosphate or phosphonate ester zwitterionic group.

13. A process according to claim 12 in which X is a group of the general formula II



II

in which the moieties  $A^3$  and  $A^4$ , which are the same or different, are -O-, -S-, -NH- or a valence bond, preferably -O-, and  $W^+$  is a group comprising an ammonium, phosphonium or sulphonium cationic group and a group linking the anionic and cationic moieties which is preferably a  $C_{1-12}$ -alkanediyl group,

preferably in which  $W^+$  is a group of formula  $-W^1-N^+R^3_3$ ,  $-W^1-P^+R^4_3$ ,  $-W^1-S^+R^4_2$  or  $-W^1\text{-Het}^+$  in which:

$W^1$  is alkanediyl of 1 or more, preferably 2-6 carbon atoms optionally containing one or more ethylenically unsaturated double or triple bonds, disubstituted-aryl (arylene), alkylene arylene, arylene alkylene, or alkylene aryl alkylene, cycloalkanediyl, alkylene cycloalkyl, cycloalkyl alkylene or alkylene cycloalkyl alkylene, which group  $W^1$  optionally contains one or more fluorine substituents and/or one or more functional groups; and

either the groups  $R^3$  are the same or different and each is hydrogen or alkyl of 1 to 4 carbon atoms, preferably methyl, or aryl, such as phenyl, or two of the groups  $R^3$  together with the nitrogen atom to which they are attached form an aliphatic heterocyclic ring containing from 5 to 7 atoms, or the three groups  $R^3$  together with the nitrogen atom to which they are

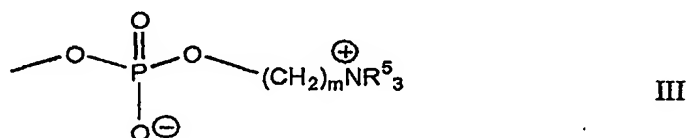
attached form a fused ring structure containing from 5 to 7 atoms in each ring, and optionally one or more of the groups  $R^3$  is substituted by a hydrophilic functional group, and

the groups  $R^4$  are the same or different and each is  $R^3$  or a group  $OR^3$ , where  $R^3$  is as defined above; or

Het is an aromatic nitrogen-, phosphorus- or sulphur-, preferably nitrogen-, containing ring, for example pyridine.

14. A process according to claim 13 in which X is a group of general formula III

10



where the groups  $R^5$  are the same or different and each is hydrogen or  $C_{1-4}$  alkyl, and m is from 1 to 4, in which preferably the groups  $R^5$  are the same preferably methyl.

15. A process according to any preceding claim in which Y is  $H_2C=CR-CO-A-$  in which R is hydrogen or methyl and A is O.

16. A polymerisation process according to any preceding claim in which B is a straight chain  $C_{2-6}$ -alkanediyl.

17. A polymerisation process according to claim 1 in which the zwitterionic monomer is 2-methacryloyloxyethyl-2'-trimethylammonium ethyl phosphate inner salt.

18. A polymerisation process according to any preceding claim in which the polymerisation mixture contains a non-polymerisable solvent, preferably in an amount, in the range of 10 to 500% by weight based on the weight of ethylenically unsaturated monomer.

19. A polymerisation process according to any preceding claim in which the ethylenically unsaturated monomer includes at least one comonomer, preferably selected from anionic, cationic and non-ionic monomers, more preferably comprising a non-ionic monomer.

20. A polymerisation process according to any preceding claim in which the catalyst comprises a transition metal compound and a ligand, in which the transition metal compound is capable of participating in a redox cycle with the initiator and dormant polymer chain, and the ligand is either  
 5 any N-, O-, P- or S- containing compound which can coordinate with the transition metal atom in a  $\sigma$ -bond, or any carbon-containing compound which can coordinate with the transition metal in a  $\pi$ -bond, such that direct bonds between the transition metal and growing polymer radicals are not formed.

21. A polymerisation process according to claim 20 in which the  
 10 transition metal compound  $M_t^{n+}X'_n$ , where:

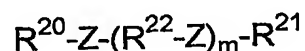
$M_t^{n+}$  may be selected from the group consisting of  $Cu^{1+}$ ,  $Cu^{2+}$ ,  $Fe^{2+}$ ,  $Fe^{3+}$ ,  $Ru^{2+}$ ,  $Ru^{3+}$ ,  $Cr^{2+}$ ,  $Cr^{3+}$ ,  $Mo^{2+}$ ,  $Mo^{3+}$ ,  $W^{2+}$ ,  $W^{3+}$ ,  $Mn^{2+}$ ,  $Mn^{3+}$ ,  $Mn^{4+}$ ,  $Rh^{3+}$ ,  $Rh^{4+}$ ,  $Re^{2+}$ ,  $Re^{3+}$ ,  $Co^{+}$ ,  $Co^{2+}$ ,  $Co^{3+}$ ,  $V^{2+}$ ,  $V^{3+}$ ,  $Zn^{+}$ ,  $Zn^{2+}$ ,  $Ni^{2+}$ ,  $Ni^{3+}$ ,  $Au^{+}$ ,  $Au^{2+}$ ,  $Ag^{+}$  and  $Ag^{2+}$ ;

15  $X'$  is selected from the group consisting of halogen,  $C_{12}$ - $C_6$ -alkoxy,  $(SO_4)_{1/2}$ ,  $(PO_4)_{1/3}$ ,  $(R^{18}PO_4)^{1/2}$ ,  $(R^{18}_2PO_4)$ , triflate, hexafluorophosphate, methanesulphonate, arylsulphonate, CN and  $R^{19}CO_2$ , where  $R^{18}$  is aryl or a straight or branched  $C_{1-20}$  alkyl and  $R^{19}$  is H or a straight or branched  $C_1$ - $C_6$  alkyl group which may be substituted from 1 to 5 times with a halogen; and  
 20  $n$  is the formal charge on the metal ( $0 \leq n \leq 7$ ).

22. A polymerisation process according to claim 12 in which the metal compound is  $CuHal$  or  $RuHal_2$  where Hal is chlorine or bromine.

23. A polymerisation process according to any of claims 20 to 22 wherein said ligand is selected from the group consisting of:

25 a) compounds of the formulas:



30 where:

$R^{20}$  and  $R^{21}$  are independently selected from the group consisting of H,  $C_1$ - $C_{20}$  alkyl, aryl, heterocyclyl and  $C_1$ - $C_6$  alkoxy,  $C_1$ - $C_4$  dialkylamino,

$C(=O)R^{22}$ ,  $C(=O)R^{23}R^{24}$  and  $A^7C(=O)R^{25}$ , where  $A^7$  may be  $NR^{26}$  or O;  $R^{22}$  is alkyl of from 1 to 20 carbon atoms, aryloxy or heterocyclyloxy;  $R^{23}$  and  $R^{24}$  are independently H or alkyl of from 1 to 20 carbon atoms or  $R^{23}$  and  $R^{24}$  may be joined together to form an alkanediyl group of from 2 to 5 carbon atoms, thus forming a 3- to 6-membered ring;  $R^{25}$  is H, straight or branched  $C_1$ - $C_{20}$  alkyl or aryl and  $R^{26}$  is hydrogen, straight or branched;  $C_1$ - $C_{20}$ -alkyl or aryl; or  $R^{20}$  and  $R^{21}$  may be joined to form together with Z, a saturated or unsaturated ring;

Z is O, S,  $NR^{27}$  or  $PR^{27}$ , where  $R^{27}$  is selected from the same group as  $R^{20}$  and  $R^{21}$ , and where Z is  $PR^{27}$ ,  $R^{27}$  can also  $C_1$ - $C_{20}$  alkoxy or Z may be a bond  $CH_2$  or a fused ring, where one or both of  $R^{20}$  and  $R^{23}$  is heterocyclyl, each  $R^{22}$  is independently a divalent group selected from the group consisting of  $C_1$ - $C_8$  cycloalkanediyl,  $C_1$ - $C_8$  cycloalkanediyl, arenediyl and heterocyclylene where the covalent bonds to each Z are at vicinal positions or  $R^{22}$  may be joined to one or both of  $R^{20}$  and  $R^{21}$  to formulate a heterocyclic ring system; and

m is from 1 to 6;

b) CO;

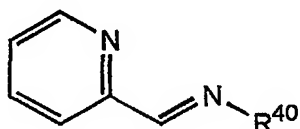
c) porphyrins and porphycenes, which may be substituted with from 1 to 6 halogen atoms,  $C_{1-6}$  alkyl groups,  $C_{1-6}$ -alkoxy groups,  $C_{1-6}$  alkoxy carbonyl, aryl groups, heterocyclyl groups, and  $C_{1-6}$  alkyl groups further substituted with from 1 to 3 halogens;

d) compounds of the formula  $R^{23}R^{24}C(C(=O)R^{25})_2$ , where  $R^{25}$  is  $C_{1-20}$  alkyl,  $C_{1-20}$  alkoxy, aryloxy or heterocyclyloxy; and each of  $R^{23}$  and  $R^{24}$  is independently selected from the group consisting of H, halogen,  $C_{1-20}$  alkyl, aryl and heterocyclyl, and  $R^{23}$  and  $R^{24}$  may be joined to form a  $C_{1-8}$  cycloalkyl ring or a hydrogenated aromatic or heterocyclic ring, of which the ring atoms may be further substituted with 1 to 5  $C_{1-6}$  alkyl groups,  $C_{1-6}$  alkoxy groups, halogen atoms, aryl groups, or combinations thereof; and

e) arenes and cyclopentadienyl ligands, where said cyclopentadienyl ligand may be substituted with from one to five methyl

groups, or may be linked through and ethylene or propylene chain to a second cyclopentadienyl ligand.

24. A polymerisation process according to claim 23 in which the ligand is bipyridine, triphenylphosphine, 1,1,4,7,10,10-hexamethyl-  
5 triethylene tetramine, or a compound of the general formula VII

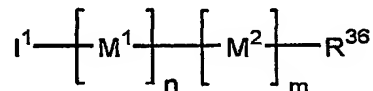


VII

- 10 where  $R^{40}$  is an alkyl or substituted alkyl group, in which the substituent is selected from amino, including alkylamino and acylamino, alkoxy, hydroxy, acyl, acyloxy, alkoxycarbonyl, heterocyclyl, ionic, and halogen substituents.

25. A compound comprising a conjugate of a biologically active moiety and a polymeric group having a general formula:

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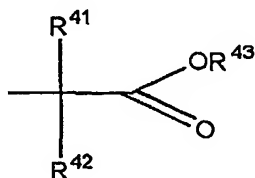


VIII

- in which  $M^1$  is the divalent group formed when the compound of the general formula I as defined in claim 1 is polymerised,  $M^2$  is the divalent group  
20 formed when a comonomer as defined in claim 19 is polymerised, and  $I^1$  is the residue of the initiator defined in claim 1 which comprises said biologically active moiety, and  $R^{36}$  is a monofunctional group or atom which terminates the polymeric group  $M^1_n$ ,  $n$  is at least 2 and  $m$  is at least 0.

26. A compound according to claim 25 in which  $I^1$  is  $-CR^{11}R^{12}R^{13}$  in  
25 which  $R^{11}$  to  $R^{13}$  are as defined in claim 2.

27. A compound according to claim 25 in which  $I^1$  is a group



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in which  $R^{41}$ ,  $R^{42}$  and  $R^{43}$  are as defined in any of claims 3 to 9.

28. A compound according to any of claims 25 to 27 in which the compound has a molecular weight in the range 1000 to 100,000, preferably 20,000 to 50,000.

29. A compound according to any of claims 25 to 28 which has a polydispersity less than 1.5, preferably less than 1.2.

30. A compound according to any of claims 25 to 29 in which  $M^1$  is the divalent group formed when a compound of the general formula I as defined in any of claims 12 to 17 is polymerised.

31. A compound according to claim 25 in which groups  $M^1$  and  $M^2$  are randomly arranged.

32. A compound according to claim 25 in which the polymeric group is a block polymeric group, in which one block comprises residues  $M^1$  and another block comprises residues  $M^2$ , and in which either the  $M^1$ -containing block, or the  $M^2$  containing block is attached to  $I^1$ .

33. A compound according to any of claims 25 to 32 which is soluble in water.

**ABSTRACTS****POLYMER CONJUGATES**

The present invention relates to conjugates of biologically active compounds, preferably therapeutically active compounds, with polymeric moieties having low polydispersity, as well as controlled polymerisation processes for producing the conjugates. An initiation for a controlled radical polymerisation process comprises a biologically active, usually therapeutically active, moiety and the monomer includes zwitterionic monomer for instance 2-methacryloyloxyethyl-2'-trimethylammonium ethyl phosphate inner salt. The process allows close control of the molecular weight and polydispersity of the polymeric moiety and the possibility of optimising the delivery characteristics of the active agent.



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